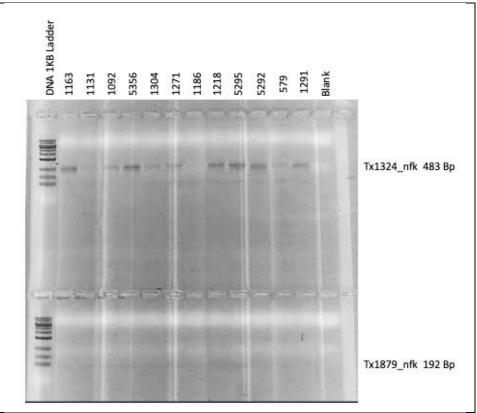
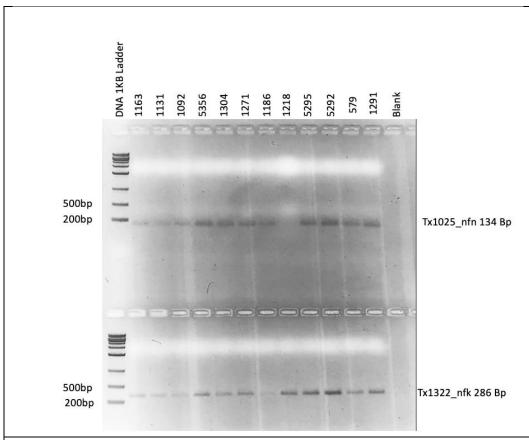
Supplementary Figures

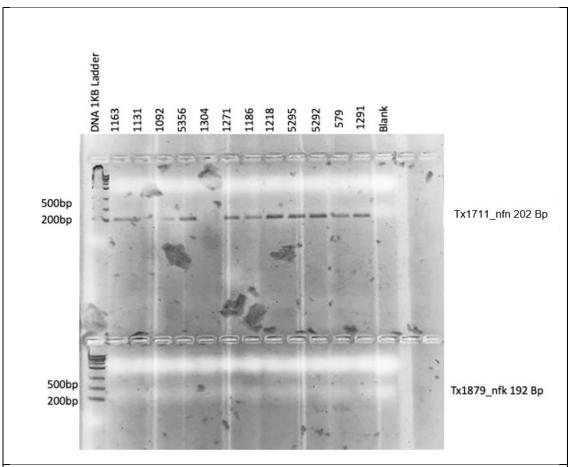


Supplementary Figure 1: PCR validation for new high confidence transcripts BambuTx1324 (*NELFA*) and BambuTx1879 (*MAOB*). Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx1324 is a new RNA isoform from the *NELFA* gene and was succesfully validated. BambuTx1879 is a new RNA isoform for the *MAOB* gene and was not validated.



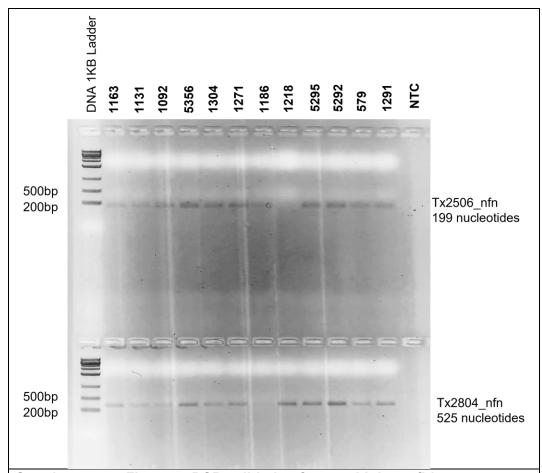
Supplementary Figure 2: PCR validation for new high confidence transcripts BambuTx1025 (New gene body) and BambuTx1322 (*SLC26A1*).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx1025 is a new RNA isoform from a newly discovered gene body and was succesfully validated. BambuTx1322 is a new RNA isoform for the *SLC26A1* gene and was succesfully validated.



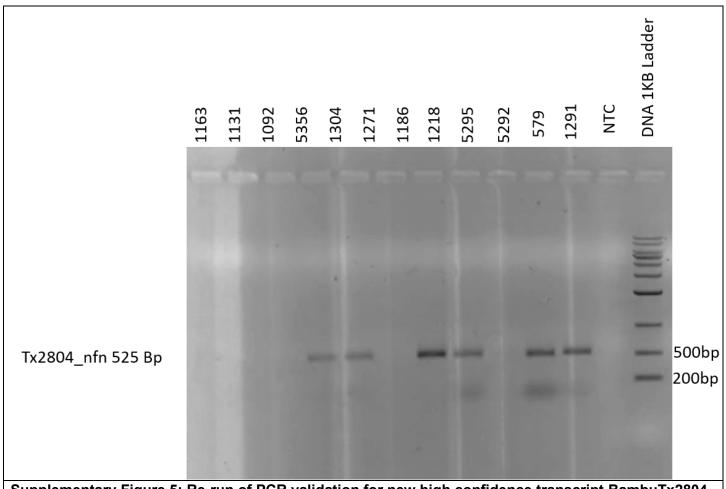
Supplementary Figure 3: PCR validation for new high confidence transcripts BambuTx1711 (New gene body) and BambuTx1879 (*MAOB*).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx1711 is a new RNA isoform from a newly discovered gene body and was successfully validated. BambuTx1879 is a new RNA isoform for the *MAOB* gene and was not validated.



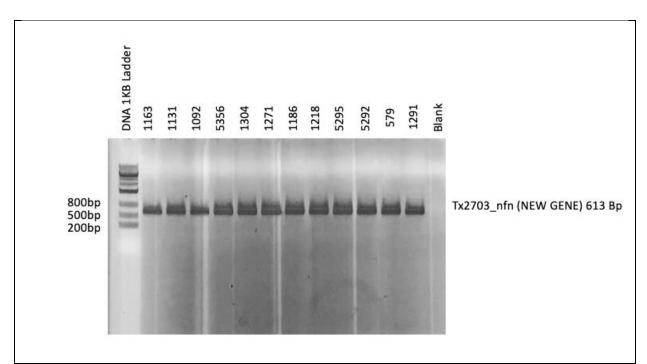
Supplementary Figure 4: PCR validation for new high confidence transcripts BambuTx2506 (New gene body) and BambuTx2804 (New gene body).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx2506 is a new RNA isoform from a new gene body and was successfully validated. BambuTx2804 is a new RNA isoform from a new gene body and was not validated in this figure. We did not consider the BambuTx2804 validation successful on this gel because the band is considerably smaller than the predicted 525 nucleotide PCR product. However, we ran another gel and successfully validated it in **Supplementary Figure 5**. That PCR reaction was ran with a higher annealing temperature to prevent formation of primer dimers.



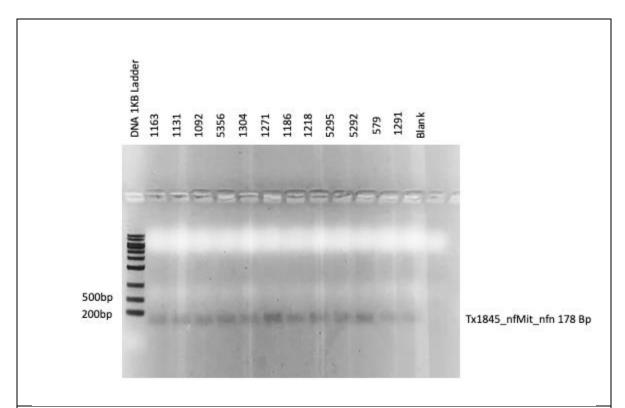
Supplementary Figure 5: Re-run of PCR validation for new high confidence transcript BambuTx2804 (New gene body).

Number above the lanes is the sample id for the 12 samples used in this study. Label on the left side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx2804 is a new RNA isoform from a new gene body and was successfully validated.



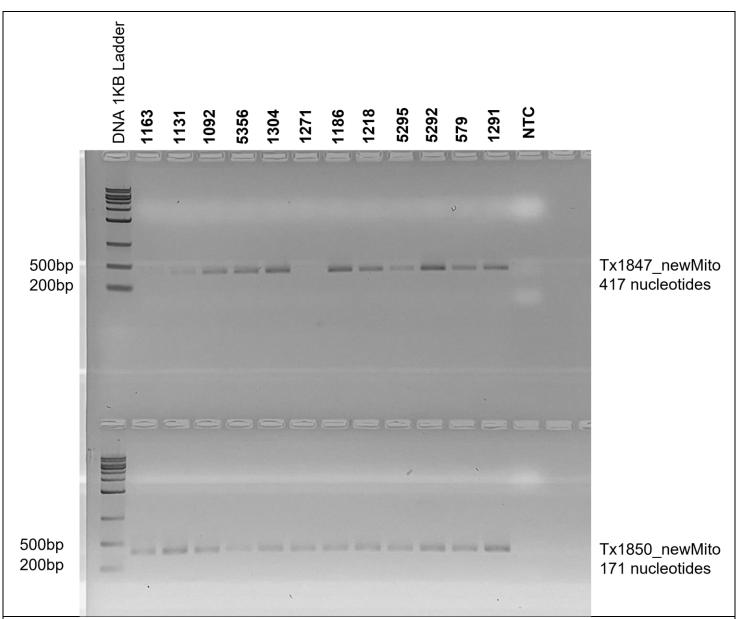
Supplementary Figure 6: PCR validation for new high confidence transcripts BambuTx2703 (New gene body).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx2703 is a new RNA isoform from a newly discovered gene body and was succesfully validated.



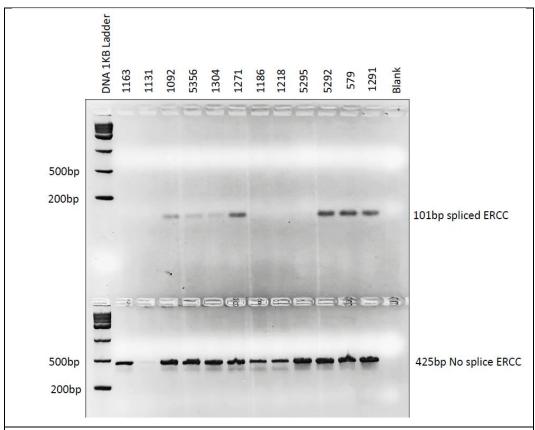
Supplementary Figure 7: PCR validation for new high confidence transcripts BambuTx1845 (New spliced mitochondrial isoform).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx1845 is a new spliced RNA isoform from mitochondria and was successfully validated.



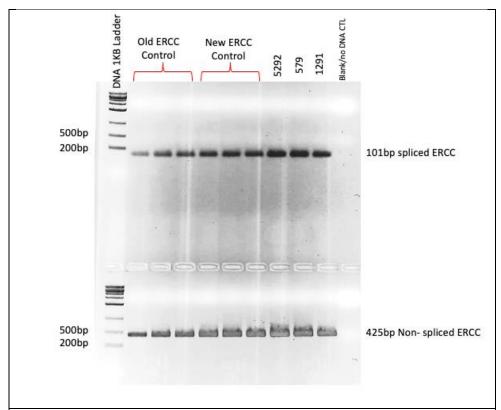
Supplementary Figure 8: PCR validation for new high confidence transcripts BambuTx1847 and BambuTx1850 (New spliced mitochondrial isoforms).

Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the new high-confidence RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. BambuTx1847 is a new spliced RNA isoform from mitochondria and was successfully validated. BambuTx1850 is a new spliced RNA isoform from the mitochondria and was not validated. We did not consider the BambuTx1850 validation successful on this gel because the band is considerably larger than the predicted 171 nucleotide PCR product.



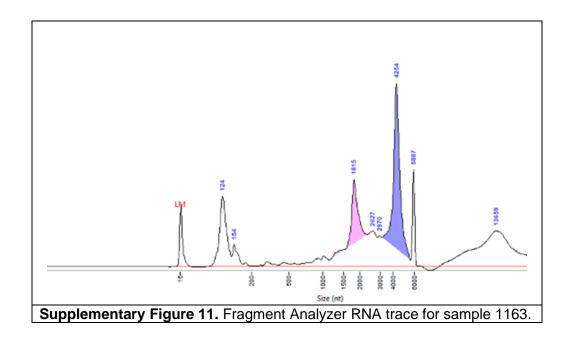
Supplementary Figure 9: PCR validation for spliced ERCC RNA isoform in 12 samples.

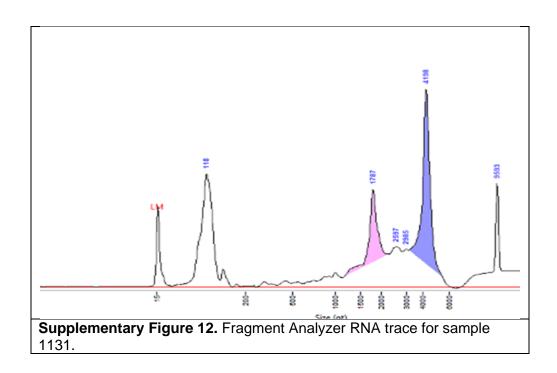
Number above the lanes is the sample id for the 12 samples used in this study. Labels on the right side of the figure indicate transcript id for the RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. ERCC spliced refers to the new spliced ERCC RNA isoform found in our data, which was successfully validated. No splice ERCC refers to the regular ERCC sequence that is no spliced, which was also validated successfully, as expected.

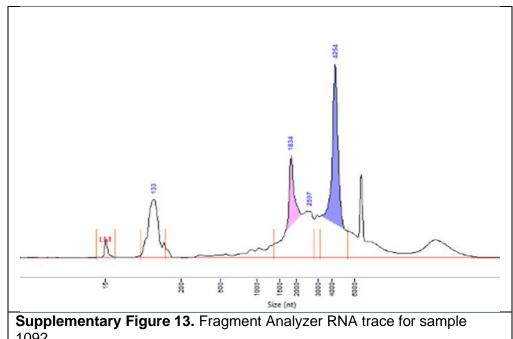


Supplementary Figure 10: PCR validation for spliced ERCC RNA isoform in two different ERCC batches and 4 samples.

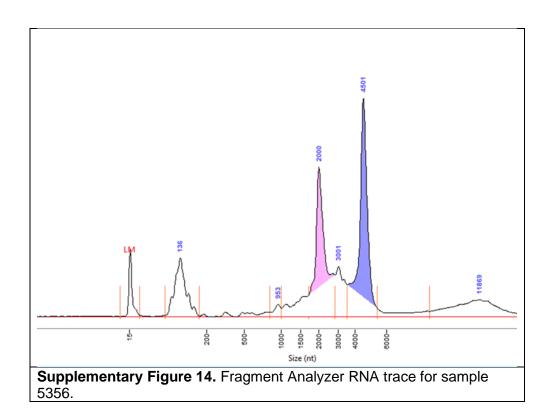
Number above the lanes is the sample id for 4 samples used in this study, old ERCC control refers to an older ERCC batch whereas new ERCC refers to a fresh ERCC batch we tested with. Labels on the right side of the figure indicate transcript id for the RNA isoform being validated and the expected product length from the PCR primers used to amplify the RNA isoform. ERCC spliced refers to the new spliced ERCC RNA isoform found in our data, which was successfully validated. No splice ERCC refers to the regular ERCC sequence that is no spliced, which was also validated successfully as expected.

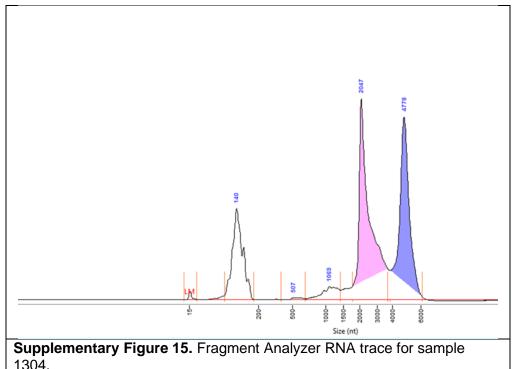




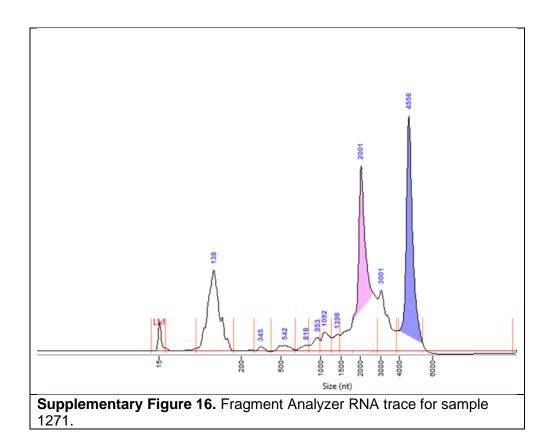


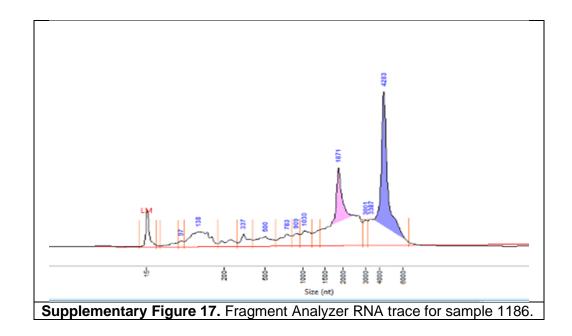
1092.

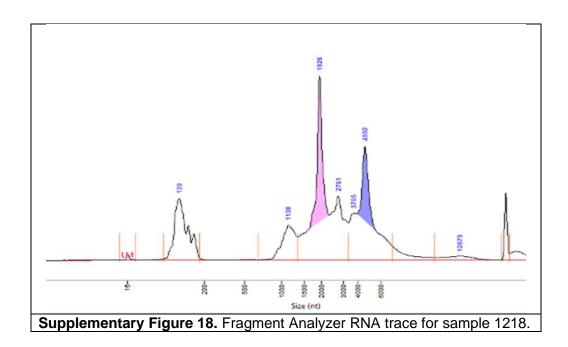


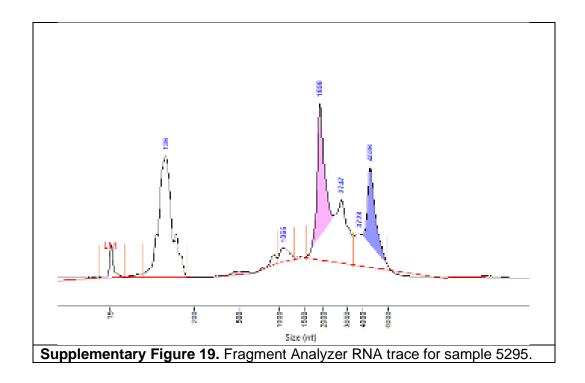


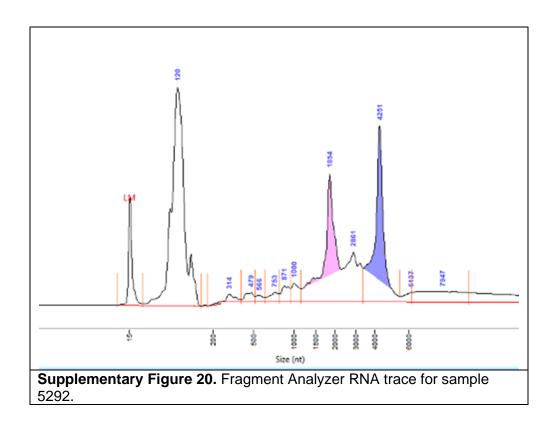
1304.

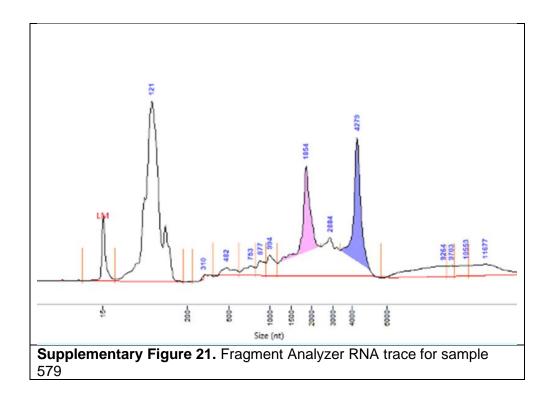




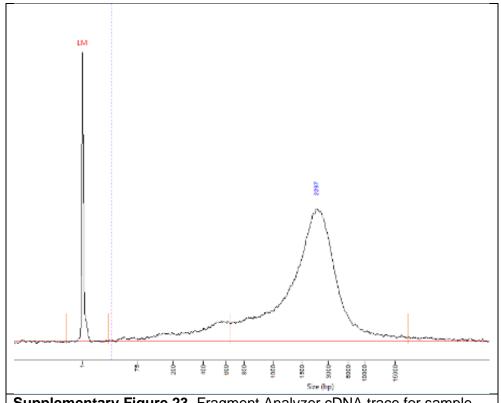




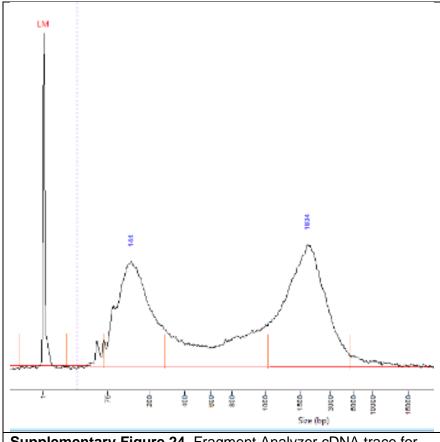




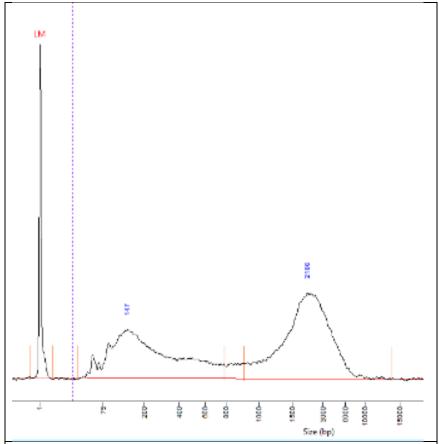
Supplementary Figure 22. Fragment Analyzer RNA trace for sample 1291



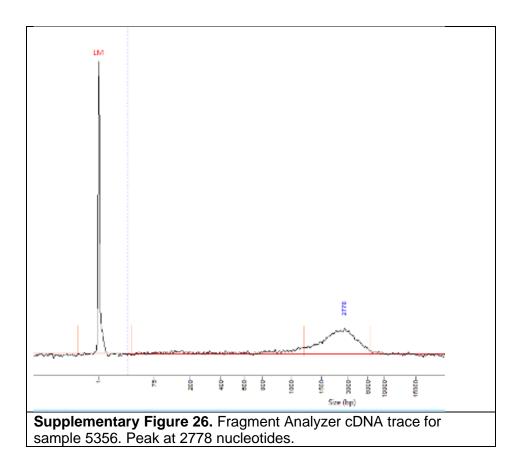
Supplementary Figure 23. Fragment Analyzer cDNA trace for sample 1163. Peak at 2297 nucleotides.

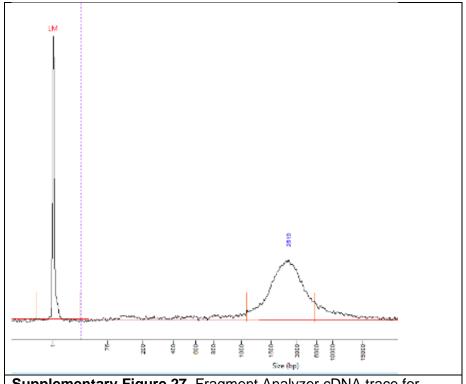


Supplementary Figure 24. Fragment Analyzer cDNA trace for sample 1131. Lower peak at 144 nucleotides, higher peak at 1834 nucleotides

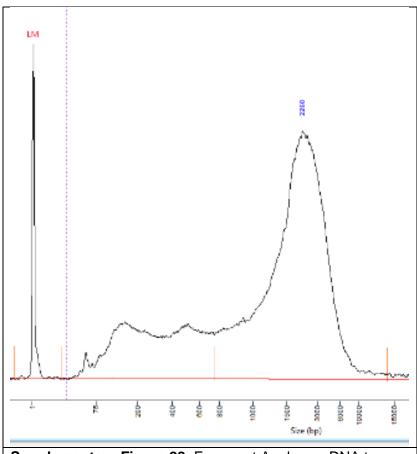


Supplementary Figure 25. Fragment Analyzer cDNA trace for sample 1092. Lower peak at 147 nucleotides, higher peak at 2186 nucleotides

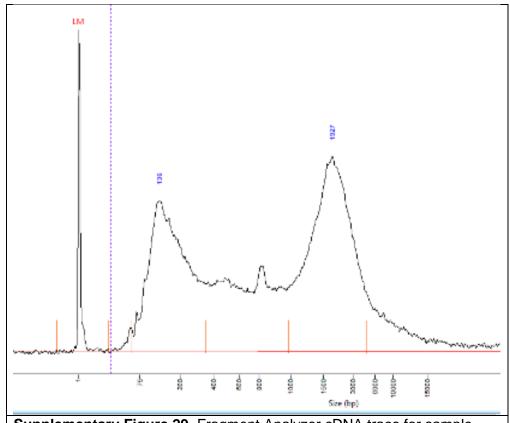




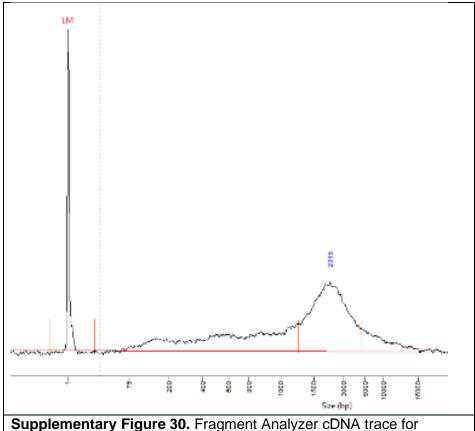
Supplementary Figure 27. Fragment Analyzer cDNA trace for sample 1304. Peak at 2519 nucleotides.



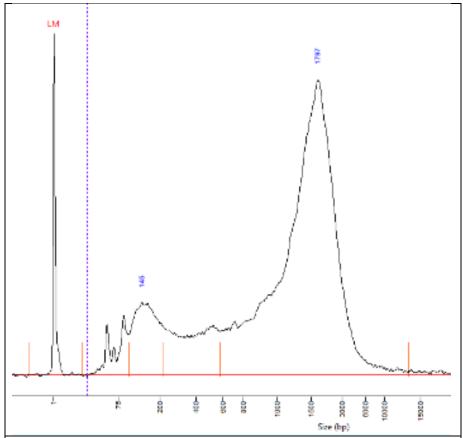
Supplementary Figure 28. Fragment Analyzer cDNA trace for sample 1271. Peak at 2260 nucleotides.



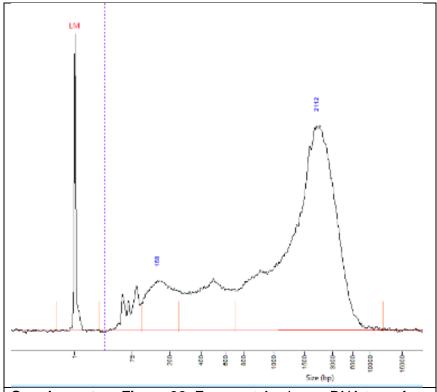
Supplementary Figure 29. Fragment Analyzer cDNA trace for sample 1186. Lower peak at 136 nucleotides, higher peak at 1927 nucleotides.



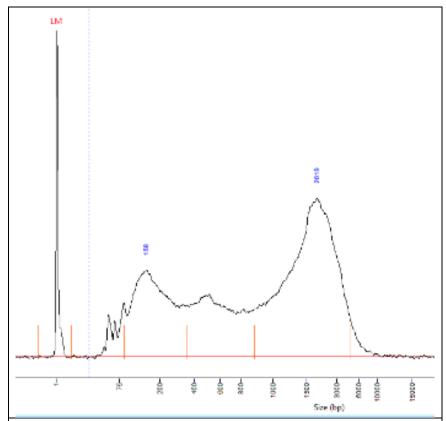
Supplementary Figure 30. Fragment Analyzer cDNA trace for sample 1218. Peak at 2315 nucleotides.



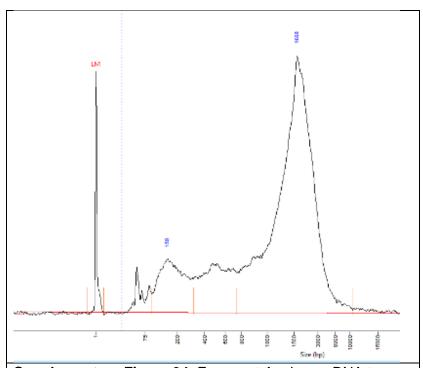
Supplementary Figure 31. Fragment Analyzer cDNA trace for sample 5295. Lower peak at 145 nucleotides, higher peak at 1797 nucleotides.



Supplementary Figure 32. Fragment Analyzer cDNA trace for sample 5292. Lower peak at 158 nucleotides, higher peak at 2112 nucleotides.



Supplementary Figure 33. Fragment Analyzer cDNA trace for sample 579. Lower peak at 158 nucleotides, higher peak at 2019 nucleotides.



Supplementary Figure 34. Fragment Analyzer cDNA trace for sample 1291. Lower peak at 159 nucleotides, higher peak at 1668 nucleotides.